

A Comparison of the Microbial Count of Washed and Unwashed Oranges after Storage at Room and Refrigeration Temperature Conditions

Ahmad Allafi¹, Saad Busamri²

1 Assistant Professor, Department of Family Sciences, College for Women, Kuwait University, P.O.Box: 5969, Safat 13060, Kuwait. Tel: (965)2498-3161, Fax: (965)2251-3929
ahmadallafi@yahoo.com

2 Lecturer, Nutrition and Food Science Department, The Public Authority for Applied Education & Training, Kuwait

ABSTRACT:

Most people believe that fresh fruits and vegetables that are meant to be peeled need not be washed right before eating as the rind acts as a protective covering and isn't removed until eaten. The main problem with this belief is that if the rind isn't washed properly before peeling, the microbial load from the peel ends up being transferred to the flesh. The significance of washing oranges prior to peeling was examined in this study. By storing the oranges at two different (room and refrigerator) temperatures, the rate of the microbial growth was also studied. Homogenization of the oranges was done by stomaching (peeled oranges) and by the use of an orbital shaker (unpeeled oranges). A series of dilutions for each sample was done by plating it separately on culture plates containing acidified potato dextrose agar (PDA) and tryptic soy agar (TSA). The findings of this study indicated that washing oranges prior to peeling decreased the microbial load. However, the storage temperature had little effect, if any, on the overall microbial counts of the samples.

Keywords: orange, washed, unwashed, refrigeration

INTRODUCTION

Oranges (*Citrus sinensis*) belong to the rue family (Rutaceae) and come originally from Southern China. In general, the different varieties of oranges like other citrus fruits, thrive in semitropical regions such as Florida and subtropical regions such as California and the Mediterranean. It also grows in tropical areas, mainly parts of South and Central America as well as some areas of Southeast Asia. The harvest yield of the tropical fruit is less predictable than from the semitropical areas because, the hot weather enhances the rate of maturation of the fruit. In addition, citrus fruits from the tropical areas often possess green-colored rinds because low temperature during the nights are necessary for the bright color to set [1].

Survey of raw fruits and vegetables demonstrates that there are potentials for wide range of these products becoming contaminated with microorganism, especially the pathogenic ones [2]. Fresh produce may contain plant pathogenic and spoilage-microorganisms that would decrease the shelf-life and acceptability of fresh produce. Microbes responsible for fruits and vegetables decay include bacteria (*Lactobacillus* spp.), yeast (*Saccharomyces* spp.), and molds (*Rhizopus* spp.). These organisms can render the fresh fruits and vegetables making it unfit for human consumption; by causing their deterioration and leading to reduction in quality, texture, off flavor development and loss of nutrients [3].

Peel of citrus fruits is a natural barrier to most microbial contaminations. Surface molds will eventually cause the fruit to decompose. Once a fruit is broken, microbial decay can occur rapidly [4].

International Journal of BioSciences, Agriculture and Technology (2012), Volume 4, Issue 1, Page(s):1-3

Microbial contamination of produce can be attributed to many factors, including variety and maturity, field practices (e.g. irrigation, fertilizers, handling by workers), contaminated bins and equipments, and transport vehicles [5, 6, 7]. Several possible sources of contamination that would result in the spoilage of oranges may occur at different stages such as during the growth, the handling processes and after distribution. During the growth period, the soil, insects, and animals could be the sources of contamination. The water used for irrigation or washing during the processing may contain pollutants that would result in the contamination of the oranges when sprayed. Post-harvest losses in citrus fruits have been observed too much due to which the fruit loses its quality [8]. Packing, storage, and shipping operations with improper sanitation can contribute microorganisms to proliferate rapidly [9]. Improper handling of oranges by employees and customers at the store may also contribute to the contamination. This may result in the decaying of the fruit to such a point that it would turn unappealing to eat. Many fruits, such as oranges, have rinds or peels that can keep many microorganisms from infesting and proliferating. It has been documented however, that those microbes can be transferred from the rind to the inner flesh, which could be harmful to the individual eating the spoiled fruit. The standard levels of microorganisms that are considered acceptable for fruits are $10^3 - 10^5$ CFU/g [10].

Since the rind isn't removed until right before eating, most people believe that these types of fruits do not need to be washed. The main problem with this belief is that if the rind is not washed properly before peeling,

the microbial load from the peel ends up being transferred to the flesh. Since the product is susceptible to all different types of contamination, one is exposed to high risk of infection. The usual practice of washing of citrus fruits after harvest was an effective method for removing spores from the surfaces of fruits [11]. Therefore, the objectives of this study were: (1) To determine if washing an orange before peeling would decrease its total microbial load. (2) To find out if microbes are transferred to the flesh after peeling. (3) To verify whether storage temperatures affect the microbial count of an orange.

MATERIALS AND METHODS

A total of eight oranges were split into two groups of four based on their storage temperature - refrigerated and room temperature. These two groups were then further split and one half were rinsed with tap water with minimal friction, while the other half remained unwashed. Of the washed oranges, one was peeled while the other remained intact. The same was applied to the unwashed oranges as well. The individual samples were as follows: refrigerated, washed, peeled; refrigerated, washed unpeeled; refrigerated, unwashed, peeled; refrigerated, unwashed, unpeeled; room temperature, washed, peeled; room temperature, washed, unpeeled; room temperature, unwashed, peeled; and room temperature, unwashed, unpeeled.

All oranges that remained intact, that is, unpeeled were then shaken on an orbital shaker in 99 ml of peptone water for 2 minutes. Whereas, oranges that were peeled were stomached in 99 ml of peptone water for 2 minutes. A dilution series was then performed on each; 10^{-2} , 10^{-3} , 10^{-4} for those that were peeled and 10^{-1} , 10^{-2} , and 10^{-3} for those that remained unpeeled in 9 mL peptone blanks.

0.1 mL of each of the last three dilutions was then double-spread plated on both the acidified PDA and the TSA as to gain a better average for the total growth of the microbes. A total of 96 plates were then incubated at 30°C for 5 days to maximize the growth of yeasts and molds which grow at a fairly slow rate. After this incubation period, each plate was enumerated to determine the CFU/orange.

RESULTS AND DISCUSSION

With reference to the first objective, consumers mistakenly assume that fruits and vegetables that are peeled just before consumption is safe to eat without washing due to the removal of the outer layer. The current study has proved that washing generally decreases the microbial load. There were a few instances in our results, where the washed samples did contain higher counts, as shown in Table 1. However, these discrepancies were thought to be due to the observed mold growth that was already present on two of the washed samples. Also, another possible cause might have been due to the use of tap water instead of

distilled water to rinse the oranges since microbes could be present in unsterilized water.

According to Table 2, the samples that were peeled displayed high microbial counts. These higher counts indicate that some of the microbial load from the rind was transferred to the flesh during the peeling process. One also has to take into account that three of the four samples that were peeled contained rot. It was expected and verified that this would result in a higher count on and in the flesh due to the higher concentration of microbes on the rind. This hypothesis was confirmed due to the observation of similar colonies on the plated samples of both the peeled and the unpeeled oranges. For example, almost all plates contained fuzzy green colonies presumably tested to be *Penicillium* spp.

Table 1. A comparison of the microbial counts of yeast and mold (and possibly acid tolerant bacteria) in samples of washed^a vs. unwashed oranges.

Orange Conditions	Average Count (CFU/orange) ^b	
	Acidified PDA ^c	TSA ^d
Washed	3.8×10^3	1.2×10^4
	4.8×10^3	4.1×10^3
	2.3×10^3	1.9×10^3
	9.8×10^3	6.7×10^3
Unwashed Oranges	1.4×10^4	2.0×10^4
	9.7×10^3	2.3×10^4
	9.7×10^3	7.0×10^3
	5.5×10^3 (est.) ^e	4.5×10^3 (est.)

^aRinsed with tap water with minimal friction.

^bResults are presented as colony forming unit (CFU)/orange.

^cPotato dextrose agar.

^dTryptic soy agar.

^eNo sampled plate counts in acceptable countable range.

Table 2. A comparison of the microbial counts of yeast and mold (and possibly acid tolerant bacteria) in samples of peeled vs. unpeeled oranges.

Orange Conditions	Average Count (CFU/orange) ^b	
	Acidified PDA ^c	TSA ^d
Peeled	2.3×10^5	1.9×10^5
	9.8×10^5	6.7×10^5
	9.7×10^5	7.0×10^5
	5.5×10^5 (est.) ^e	4.5×10^5 (est.)
Unpeeled	3.8×10^3	1.2×10^4
	4.8×10^3	4.1×10^3
	1.4×10^4	2.0×10^4
	9.7×10^3	2.3×10^4

^bResults are presented as colony forming unit (CFU)/orange.

^cPotato dextrose agar.

^dTryptic soy agar.

^eNo sampled plate counts in acceptable countable range.

Consumers have a choice as to what temperature they would want to store the fruits and vegetables. Some opt to refrigerate, while others opt to allow their product to remain at room temperature. At refrigeration temperature, fungi and some bacteria, including psychrotrophs, survive and proliferate. On the other hand, at room temperature, other bacteria and some fungi can survive and proliferate. Fungi tend to prefer lower temperatures than most bacteria, which is why they display optimal growth at refrigeration temperatures. After conducting the experiment, it was determined that storage temperature had little effect, if any, on the overall microbial counts of the samples (Table 3). This was due to counting total microbes grown as opposed to counting the fungi and bacteria

separately. However, a higher rate of mold growth was observed on the refrigerated samples plates. Many microbial contaminants are part of the environment so fruits and vegetables may be inadvertently contaminated. Raw and minimally-processed fruits and vegetables are an essential part of the people's diet all around the world. So every effort should be made to prevent contamination of fruits and vegetables during production, transport, and handling. Hence, the findings of the current study suggest that oranges along with other fruits and vegetables are to be sanitized, disinfected, or at least washed thoroughly before peeling for consumption.

Table 3. A comparison of the microbial counts of yeast and mold (and possibly acid tolerant bacteria) in orange samples stored at room temperature vs. refrigeration temperature.

Orange Conditions	Average Count (CFU/orange) ^a	
	Acidified PDA ^b	TSA ^c
Room Temperature	3.8x10 ³	1.2x10 ⁴
	2.3x10 ³	1.9x10 ⁵
	1.4x10 ⁴	2.0x10 ⁴
	9.7x10 ³	7.0x10 ⁵
Refrigeration Temperature	4.8x10 ³	4.1x10 ³
	9.8x10 ⁶	6.7x10 ⁶
	9.7x10 ³	2.3x10 ⁴
	5.5x10 ³ (est.) ^d	4.5x10 ³ (est.)

^aResults are presented as colony forming unit (CFU)/orange.

^bPotato dextrose agar.

^cTryptic soy agar.

^dNo sampled plate counts in acceptable countable range.

REFERENCES

- [1] Martin-Belloso, O. &Fortuny, R.S. (2010). *Advances in Fresh-Cut Fruits and Vegetables Processing* 1st ed. Pp. 87-144. Florida, USA: CRC Press.
- [2] Kader,A.A.(1997). Quality in Relation to Marketability of Fruits and Vegetables .Proceeding of the 5th Fruit Nut and Vegetables Eng Symposium, University of California, Davis. In: Studman, C.J.(Ed), CIGR Handbook of Agricultural Engineering, ASAE,1999,St.Joseph MI, USA,4 (3) :243-272
- [3] Obetta.S.E, Nwakonobi.T.U, and Adikwu.O.A,2011.Microbial Effects on Selected Stored Fruits and Vegetables under Ambient Conditions in Makurdi, Benue State , Nigeria. Research.J of Applied Sciences, Engg and Tech 3 (5): 393-398.
- [4] Dan.A.K,(1999). Citrus Processing: A Complete Guide , 2nd ed. Pp.155.
- [5] Brackett,R.E.and D.F. Splittstoesses.2001.Fruits and vegetables, Pp.515-520.In.F.P.Downo and K.Ito (ed).Compendium of methods for the microbial examination of foods, 4th Ed.APHA, Washington ,DC.
- [6] Bucks,J.N and R.walcott 2003.Recent trends in microbial safety of fruits and vegetables.Amer.Phytophath.Soc.Plant management Network.available at <http://www.apsnet.org/online/features/safety>.
- [7] Waller,J.M. 2002. Post harvest diseases,pp:39-54, In.J.M.Waller, J.M.Lenne and S.J.Waller (ed). Plant Pathologist's Pocketbook, Cab Intl.Oxon, UK
- [8] Tariq,M.A, F.M.Tahir, A.A.Aasif and M.A.Pervez, 2001. Effect of Controlled Atmosphere Storage on

- Damaged Citrus Fruit Quality. Int.J.Agric.Biol;3: 9-12
- [9] DeRoeve, C.(1998).Microbiological Safety Evaluations and Recommendations on Fresh Produce. Food Control,9(6) : 321-347.
- [10] Yousef, A.E., Carlstrom, C. &Yousef, A. (2002). *Food Microbiology: A Laboratory Manual* 1st ed. Pp. 27-60. New Jersey, USA: Wiley-Interscience
- [11] Tariq,M.A, F.M.Tahir, A.A.Aasif and J.Iqbal, (2001). Effect of Washing and Seal Packaging on Scuffing Damaged Citrus Fruits Quality. Int.J.Agric.Biol;3: 461-3